GLUCOSE LOAD TOLERANCE IN SPINAL MUSCULAR ATROPHY TYPE II

by

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STATEMENT OF THESES APPROVAL

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ABSTRACT

Children with spinal muscular atrophy (SMA) exhibit diminished lean body mass and increased fat mass compared to healthy peers; conditions that limit mobility, aggravate orthopedic issues, and can potentially affect glucose tolerance and insulin sensitivity. It is unclear whether changes in body composition increase the risk for developing metabolic disease such as type II diabetes in this population. The aim of this pilot study was to determine whether preadolescents with SMA type II display impaired glucose tolerance after glucose loading.

Data for 6 preadolescents (ages 7-11 years) with SMA type II were collected during an oral glucose tolerance test (OGTT). Baseline lab values were taken after an overnight fast to assess hemoglobin A1c, insulin-like growth factor 1 (IGF-1), blood glucose, insulin, glucagon, alanine, cortisol, and urinary ketones. Anthropometric measures and dual-energy x-ray absorptiometry (DEXA) scans evaluated body composition. Data from the OGTT indicate that 3 of the 6 patients exhibited impaired glucose tolerance with blood glucose levels >140 mg/dL. Based on homeostasis model assessment for insulin resistance (HOMA-IR) values, 4 of the 6 patients demonstrated insulin resistance and all 6 patients displayed hyperinsulinemia. Furthermore, DEXA analyses on all patients revealed high body fat percentages, with mean values of 71.6% ± 13.1. The pilot data provide evidence of glucose and insulin abnormalities in SMA type II patients. Screening protocols, such as an OGTT, may be beneficial in assessing and

tracking SMA patient risk for metabolic disease. This knowledge may influence dietary management of patients with SMA and provide further insight into glucose metabolism abnormalities in the population.

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INTRODUCTION

Spinal muscular atrophy (SMA) is classified as a severe neuromuscular disease causing proximal muscle weakness and paralysis by the degeneration of the alpha motor neurons in the spinal cord (1). The rate of incidence is estimated as 1 in 6,000 to 1 in 10,000 live births, making it the second most frequent autosomal recessive disease. SMA is characterized by mutations in the survival motor neuron 1 (SMN1) gene (1).

There are four different classifications of the disease (SMA types: I, II, III, and IV). Classifications are determined by age of onset and the degree of motor function achieved. SMA type I is the most common and severe form of the disease, affecting about 50% of those diagnosed. Type II affects 30% of all patients diagnosed (2-4). Children with SMA require treatment by an interdisciplinary team including specialists from a variety of disciplines: pulmonary, endocrinology, cardiology, neurology, genetics, nutrition, physical therapy, social work, and speech and language pathology (3). Children with SMA types I and II present with swallowing and gastrointestinal complications. Common problems include bulbar dysfunction, dysphagia, delayed gastric emptying, constipation, gastroesophageal reflux, aspiration, and abdominal distension/bloating. Due to swallowing difficulties and poor weight gain, many SMA children are diagnosed with failure to thrive during infancy or early childhood and require the aid of a gastrostomy tube to meet nutrition needs (5).

Nutritional studies in this patient population have primarily examined nutrient intake, body composition, and bone mineral density in SMA type I (5, 6). Nutrition-related studies in SMA type II patients are limited; however, two studies have described the feeding difficulties and low weight gain commonly found in SMA types II and III (7, 8). Dual-energy x-ray absorptiometry (DEXA) analysis has shown that body composition comparison of SMA patients to healthy peers results in higher fat mass and diminished lean mass, despite the patients being within normal ranges on the BMI growth chart (9).

Glucose metabolism abnormalities may exist in people with SMA. Patients with neuromuscular disease and severely reduced lean body mass demonstrated an increased susceptibility to episodes of hypoglycemia during fasting (10, 11). Bowerman and colleagues examined this phenomenon in a SMA mouse model and in pancreatic cells of type I SMA patients at autopsy (12). The SMA mice experienced fasting hyperglycemia, hyperglucagonemia, and glucose intolerance after an intraperitoneal glucose tolerance test (IPGTT). Pancreatic samples examined from both the mice and from human SMA type I patients revealed that the pancreatic islets exhibited an increase in the number of glucagon producing α cells and a progressive loss in insulin producing β cells (12). A more recent study by Bowerman et al., found that the survival of motor neuron (SMN) protein facilitates pancreatic development, which when defective or reduced leads to dysfunctional glucose metabolism. Mice with depletions in SMN protein, but without the SMA phenotype, experienced increased weight gain, hyperinsulinemia, and an increase in the number of pancreatic β-cells (13).

Knowledge of glucose metabolism abnormalities and body composition among SMA type II children and adolescents during fasting is medically necessary as these patients are undergoing more surgical procedures for scoliosis, extension of growth rods, contracture release, and gastrostomy placement (14, 15). Prior to surgery, anesthesia protocols often require that patients do not eat anything by mouth for at least 8 hours or longer. Fasting can become problematic and may cause serious side effects associated with glucose metabolism abnormalities including hypoglycemia, respiratory insufficiency, and a potential fatty acid metabolism disorder (16-18). Skeletal muscle is an important source of gluconeogenic amino acids during fasting (19). In healthy humans it accounts for 30 to 45% of body weight. In patients with neuromuscular disease, however, muscle mass may be reduced to 10% of body weight and increase the likelihood of hypoglycemic events during fasting or illness (10, 11).

Body composition changes occur with progression of SMA and age. As mobility decreases and metabolism slows, lean body mass becomes diminished, while an increase in fat mass occurs (5-8). Changes in body composition may limit the ability of SMA patients to adjust to simple nutritional alterations especially during times of stress such as illness, fasting, or during surgery. Given the continual advances in medical treatment for SMA patients, including surgical procedures benefiting and extending life, understanding tolerance for glucose loading and fasting would further serve to enhance their medical care. Thus, the primary goals of this study were to: 1) generate patient blood chemistry values at dispersed time intervals before, during, and after periods of controlled fasting and 2) to assess glucose tolerance during an oral glucose tolerance test (OGTT). It is unclear whether changes in body composition increase the risk for developing metabolic

disease such as type II diabetes in this population. Therefore, body composition was measured to provide further insight into the metabolic risk factors of increased fat mass in the context of SMA disease progression. Understanding potential glucose intolerances will help inform recommendations for dietary intake and nutrition management.

METHODS

Participants

Participants were recruited through a targeted campaign and collaboration with Families of SMA (FSMA). Parents of current patients in the Pediatric Motor Disorders Research Program SMA database were contacted via email if they met eligibility criteria. Inclusion criteria consisted of a genetic diagnosis of SMA 5q and a clinical diagnosis of type II SMA, with the ability to sit, but not walk independently. Exclusion criteria consisted of acute illness, use of a feeding tube for more than 50% of dietary requirements, inability to swallow safely, a medical diagnosis of diabetes (either type I or II), and daily use of an oral hypoglycemic agent or insulin therapy.

Consent was ensured by providing verbal and written explanation of the study, answering any questions or concerns, and obtaining signed parental consent. Participant assent was obtained from all children as they were all ≥ 7 years old. All participants travelled from out of state since local recruitment was unsuccessful. The study was approved by the Institutional Review Board of the University of Utah #00064793. Study funding was contributed by Families of SMA and supported by the National Center for Advancing Translational Sciences of the National Institutes of Health under Award Number 1ULTR001067.

Study Design

A clinical pilot study was conducted. Children with SMA type II participated in an OGTT and a fasting challenge test at two separate visits 8 weeks apart. Measurement outcomes included: metabolic laboratory measurements, anthropometric measures and clinical assessment, as well as whole body DEXA.

Procedures

Overall

The study included two overnight visits at the University of Utah's Center for Clinical and Translational Sciences (CCTS) located within the University of Utah Hospital. Assessment of qualification and consent was obtained at the initial visit. Baseline measures included: DEXA analysis, anthropometric measures (weight, height, tricep skin fold (TSF), and mid-arm circumference (MAC)), as well as standard vital signs. The subjects underwent an oral glucose tolerance test, during which they were given a glucose drink and then monitored for 3 hours. Lab measures were taken at the beginning and throughout the test. A fasting test was completed at the second visit at which time the subjects fasted until low blood glucose was achieved or until 20 hours passed; however, data related to this visit are not included in this study report.

Oral Glucose Tolerance Test

Subjects were checked into the CCTS as inpatients, where consent was formally obtained by study staff. Vital signs (blood pressure, % oxygen saturation, temperature, and heart rate) were collected by the nurse. The subject's weight in kg was taken with

normal clothing, without shoes or additional gear. If the subject was in a wheelchair, the weight of the chair was recorded or tared. A Norland DEXA scan for small subjects (XR-36 software version 3.3.1, Fort Atkinson, Wisconsin) was performed on each subject as well as anthropometric measurements including: height (recumbent length, arm span), tricep skin fold (TSF) using calipers (Lange Skinfold Caliper, Cambridge, Maryland), and mid-arm circumference (MAC) using a standard tape measure (cm). Anthropometric measures were performed by the dietitian or trained study staff. The subject was served a standardized meal and 100 ml of Pediasure was given as a final snack. Regular medication was taken as needed. Water or ice chips were given ad lib to ensure adequate hydration. Any remainder of the standardized meal and snack was saved on the tray to be weighed and recorded by the dietitian the following morning. Meal analysis was done using The Food Processor (version 9.1.0, and 10.5.2, 2003 and 2009 ESHA Research, Salem, Oregon) by the graduate nutrition student. On average, approximately 400 calories were consumed, consisting of 15% protein, 52% carbohydrate, and 33% fat. The participants then fasted overnight for 8 hours.

Following the fast, urine was assessed for urinary ketones by study staff using KetoStix (Bayer HealthCare, Mishawaka, Indiana). After the first void had been collected, the intravenous (IV) catheter was placed using a 20 or 22 gauge and was set up using a normal saline (NS) drip with double stop cock assembly. Baseline samples were collected by a syringe. Specifically, 6.5 ml of blood was pulled and transferred to collection tubes using a 21 gauge or larger transfer needle. Baseline labs included analysis of hemoglobin A1c, insulin-like growth factor (IGF-1), blood glucose, insulin, glucagon, alanine, and cortisol. In addition, 0.5 ml of blood was analyzed by the YSI

glucose machine (YSI 2300 STAT PLUS, YSI Incorporated, Yellow Springs, Ohio) for safety purposes following the overnight fast.

Subjects consumed a glucose drink of 1.75 g glucose per kg body weight (not to exceed 75 g). Blood samples were then collected at 30, 60, 90, 120, and 180 minutes. The samples were analyzed for blood glucose, insulin, glucagon, alanine, and cortisol. At 180 minutes, 0.5 ml blood was again collected for analysis by the YSI glucose machine for safety purposes. Values less than <60 mg/dL required another glucose sample taken 15 minutes after eating. Urinary samples were collected and tested for ketones using KetoStix test strips throughout the 3 hours.

Data Analysis

This study centered on recording time series changes in specific laboratory measurements within a rigorously selected sample of 6 SMA patients. Blood glucose and other lab values collected during the OGTT were compared to established standards from ARUP Laboratories. Anthropometric data were analyzed against Centers for Disease Control (CDC) standards and National Health and Nutrition Examination Survey (NHANES) data collected from 1999–2004. This study is exploratory and descriptive in nature and the research data collected can be used to determine dietary protocols for the management and prevention of metabolic disease.

RESULTS

Six subjects between the ages of 7 to 11 years were enrolled with an average age of 8.9 ± 1.7 years. Preadolescents were selected to identify alterations in glucose metabolism prior to the dramatic growth and development during adolescence. In addition, preadolescence is a common age group in this population for surgical procedures, especially for the treatment of scoliosis. Four of the patients selected were male and 2 were female. Five of the patients were Caucasian and 1 was Black Hispanic. One subject (A) had a feeding tube, which provided <50% of energy needs. Participant characteristics and body composition are listed in Table 1.

Blood glucose values at the 120 minute interval indicated that 3 of the 6 patients showed glucose intolerance, with blood glucose values of 155, 169, and 185 mg/dL. The average value for all subjects was 146.3 ± 27.6 mg/dL, which is considered to be just above the lower range for glucose intolerance based on the American Diabetes Association reference of 140 to 199 mg/dL (20). A value greater than or equal to 200 mg/dL is diagnostic for diabetes at the conclusion of an oral glucose tolerance test (20). Only the value from 1 subject (C) was close to that threshold with a reading of 185 mg/dL. When comparing the SMA type II patients to documented reference values (21), both groups' blood glucose values peaked at the 30 minute mark on average. Four of the SMA patients peaked at 30 minutes, while 1 peaked at 60 and 1 at 90 minutes. Two of the subjects (C, D), identical twins, had two peaks; peaking first at 30 minutes, then

dropping at 60 minutes and increasing again at the 90 minute interval and remaining elevated to 120 minutes. All 6 subjects were able to finish the glucose drink except subject (E) who drank all but 20 mL, consuming 1.55g/kg. Subject (E) experienced hypoglycemia, which occurred at 180 minutes; blood glucose dropped to 53 mg/dL. In summary, glucose values are shown in Figure 1 along with documented reference values.

Hemoglobin A1c labs collected at baseline were all within normal ranges for 5 of the 6 subjects. Subject (B), however, had a value of 5.7%, which was above the normal ARUP reference range of 4.0-5.6%. For this subject, the blood glucose value at the end of the OGTT was within the normal range at 127 mg/dL. The 3 subjects with prediabetic blood glucose values included patients A, C, and D, all of whom had normal hemoglobin A1c values.

Insulin values were above referenced fasting values for 5 of the 6 subjects, which were greater than the ARUP reference value of 3-19 (μIU/mL). Insulin values increased by over tenfold at the 30 minute interval, corresponding to the peak in glucose values at 30 minutes. The insulin values continued to climb with 4 out of the 6 subjects peaking in insulin levels at 120 minutes. One subject peaked at 60 minutes, while another peaked at 90. All 3 of the subjects considered to have glucose intolerance values at the end of the OGTT (120 min interval) had their insulin values peak at this interval as well. Normal ranges for 120 minutes are 22-79 μIU/mL. All 6 of the patients' insulin values far exceeded normal insulin ranges at the 2 hour mark with values from 244-929 μIU/mL. One subject (F), had the highest insulin value of 929 (μIU/mL) at 2 hours, but did not have glucose intolerance. Insulin ranges are shown in Figure 2.

All of the subjects had hyperinsulinemia based on the sum of insulin values collected throughout the OGTT (baseline, 30, 60, 90, and 120 minutes). Hyperinsulinemia is defined by an OGTT insulin sum of ≥300 (μIU/mL) (22, 23). The range in total insulin values for all subjects was 1066-2641 (μIU/mL) and is illustrated in Figure 3.

Homeostasis model assessment for insulin resistance (HOMA-IR) is a validated equation for denoting insulin resistance based on the calculation of both fasting glucose and insulin values (23). Four out of the 6 subjects had insulin resistance based on their HOMA-IR values. Insulin resistance characteristics are outlined in Table 2.

Body fat composition and corresponding glucose intolerance/insulin resistance are outlined in Table 3. Three out of the 6 subjects had glucose intolerance based on the results of the OGTT, with corresponding body fat percentages of 56.7, 76.1, and 82.1%. Subject B had the highest body fat percentage value of 83.4%, but did not have impaired glucose tolerance based on OGTT results. This subject did, however, have a HgbA1c of 5.7%, a level considered prediabetic. Two subjects (A, F) did not have insulin resistance based on HOMA-IR pubertal values and correspondingly had the lowest body fat percentages of the group (56.7 and 53.5%). Subject A, despite a low body fat percentage (56.7%) and normal BMI of 15.6 kg/m², had impaired glucose tolerance with a value of 169 mg/dL.

For 3 out of the 6 subjects, alanine values peaked at baseline, whereas the other 3 peaked at the 60, 90, and 120 minute intervals. Meanwhile, for 5 out of the 6 subjects, alanine values were within the normal ARUP range of 150-570 μmol/L. Only subject (C) had alanine values >570 (μmol/L) for the entire 2 hours of testing. Cortisol values

peaked at baseline in 4 out of the 6 subjects and were above the reference value of 15 (μg/dL). IGF-1 and glucagon values were both within normal limits for all 6 subjects

Table 1: Participant Characteristics and Body Composition

Characteristic	**Measure**
Male	4
Female	2
Caucasian	5
Black Hispanic	1
Age (years)	8.9 ± 1.7
Height (cm)	131.8 ± 13.8
Weight (kg)	35.6 ± 8.3
BMI (kg/m²)	20.5 ± 2.9
Lean mass (kg)	10.4 ± 6.9
Body fat (kg)	24.8 ± 4.7
Body fat %	71.6 ± 13.1
Values are means ± SD; $n = 6$	

Table 2. Insulin Resistance Characteristics

Study Subject	Fasting Glucose (mg/dL)	Fasting Insulin (µIU/mL)	HOMA-IR	Reference HOMA-IR	Insulin Resistance	Total Insulin (µIU/mL)	Hyper-insulinemia
A	99	14	3.42	≥3.82	No	1069	**Yes**
B	103	31	7.89	≥2.67[1]	**Yes**	1393	**Yes**
C	100	38	9.38	≥2.67[1]	**Yes**	2641	**Yes**
D	85	20	3.78	≥2.67[1]	**Yes**	1066	**Yes**
E	87	29	6.23	≥2.22[1]	**Yes**	2120	**Yes**
F	91	22	4.94	≥5.22	No	2637	**Yes**
HOMA-IR=Fasting insulin x Fasting glucose/405				[1]prepubertal ≤8-9 years			

HOMA-IR values based on pubertal status with prepubertal defined as ≤8-9 years (23).

Table 3. Body Composition and Glucose Metabolism

Study Subject	BMI (kg/m²)	BMI %ile	BMI Classification	Body Fat %	HgbA1c (%)	120 min Glucose (mg/dL)	HOMA-IR
A	15.6	16.4	Normal	56.7	5.3	**169**	3.42
B	18.9	90.9	Overweight	83.4	**5.7**	127	**7.89**
C	23.3	98.8	Obese	76.1	5.1	**185**	**9.38**
D	21.3	97.5	Obese	82.1	4.9	**155**	**3.78**
E	23.0	97.9	Obese	77.5	5.4	115	**6.23**
F	20.8	86.1	Overweight	53.5	4.9	127	4.94

Bold text indicates measurements outside of normal limits.

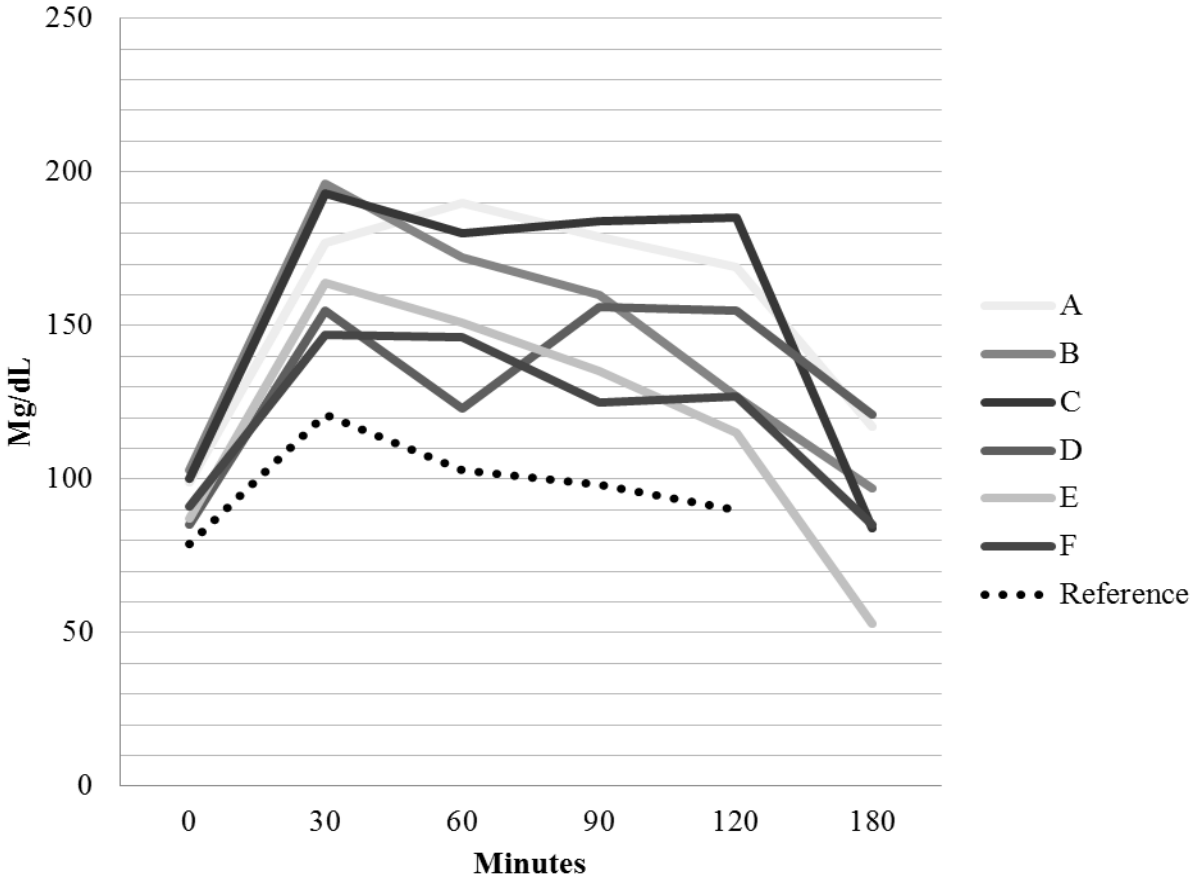

Figure 1: Glucose Ranges
Reference values are taken from Knopf et al. (21).

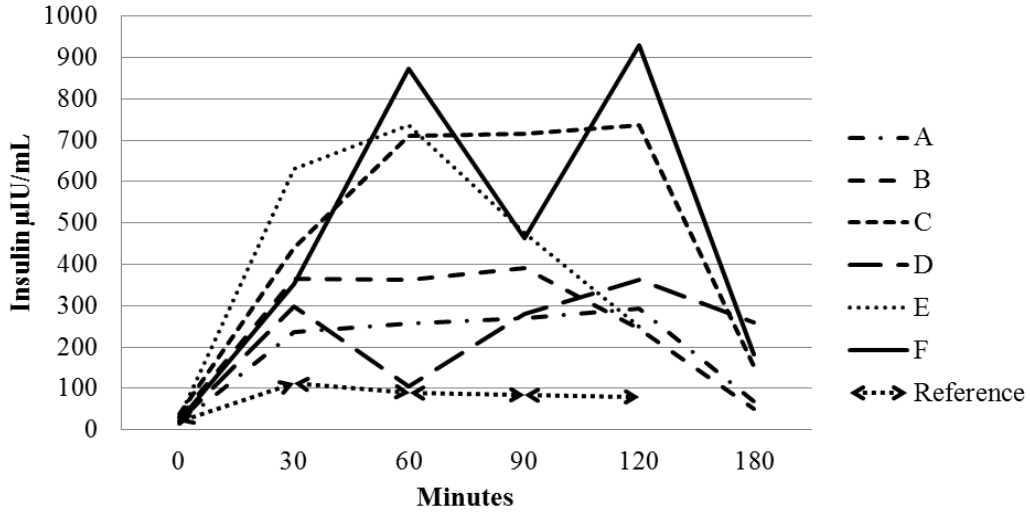

Figure 2: Insulin Ranges
Reference values from ARUP Labratories (maximum values plotted for insulin ranges during an OGTT).

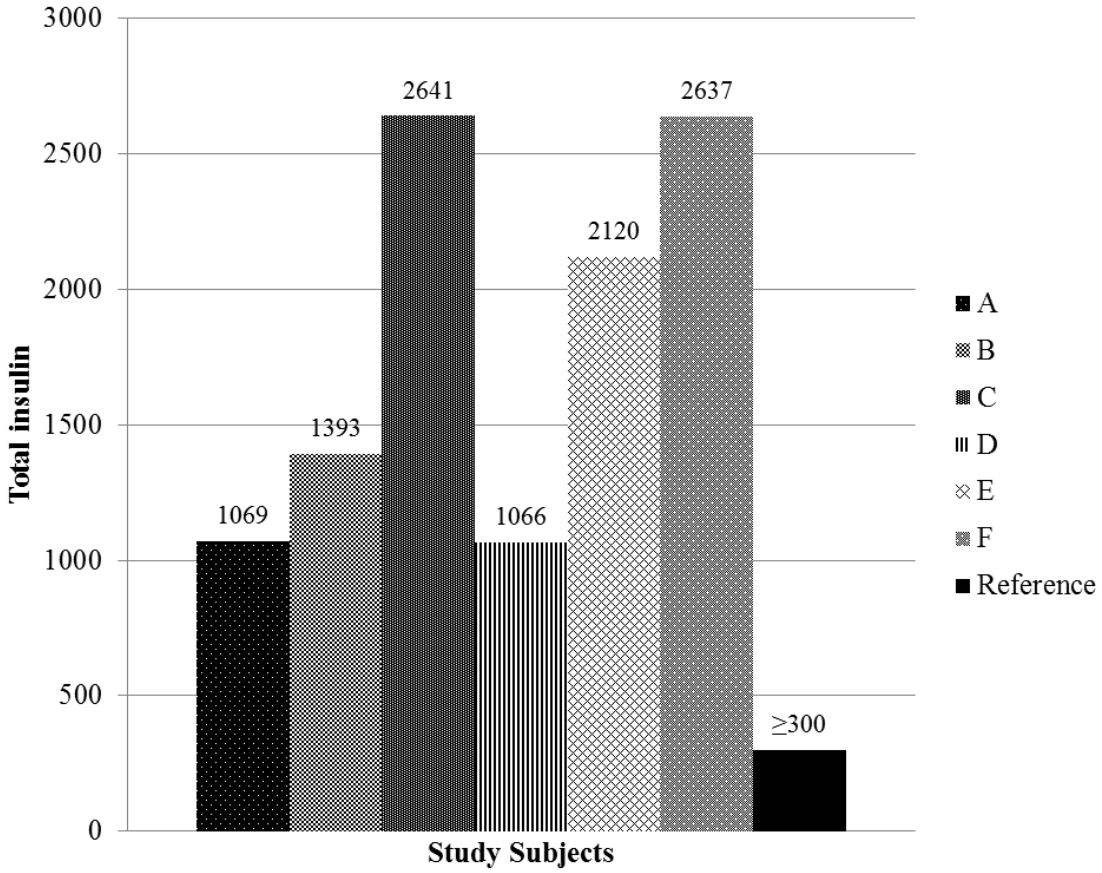

Figure 3: Hyperinsulinemia
Reference value taken from Maruhama et al. (22).

DISCUSSION

Our study discovered that 3 of the 6 enrolled subjects had glucose intolerance based on OGTT results. In addition, 4 of the 6 had insulin resistance based on HOMA-IR data. All 6 subjects had hyperinsulinemia, with total insulin sums exceeding 4 to 9 times that of the reference (≥300 (μIU/mL)). Hemoglobin Alc values were only found to be prediabetic in 1 subject; however, the subject was not found to exhibit impaired glucose tolerance at the conclusion of the OGTT. Considering that hemoglobin Alc values were within normal ranges for 5 of the 6 subjects, this test does not accurately detect glucose intolerance in SMA patients. Five of the 6 subjects were overweight/obese based on BMI values. Only 1 subject (A) fell within normal ranges on the CDC BMI chart with a BMI value of 15.6 kg/m^2. However, DEXA revealed that all subjects had an average body fat percentage greater than 70%, which suggests all of the subjects were over fat.

No other research exists for oral glucose tolerance testing in humans with SMA. Mouse models of SMA and SMN depleted mice have shown the effects of testing mice with an intraperitoneal glucose tolerance test (IPGTT) (12, 13). One mouse model tested SMA mice and found fasting hyperglycemia and hypergluconemia with diminished pancreatic β-cells. However, in the most recent study, mice with reduced SMN protein (~50% of expected) were compared to wild type mice (WT) (13). Findings revealed that despite no presence of neuromuscular disease, the SMN mice had glucose and pancreatic abnormalities. When the SMN mice were metabolically challenged with a high fat diet,

they had increased glucose levels during an IPGTT suggesting impaired glucose clearance. The reduced ability to clear glucose was attributed to an increase in abnormally located α-cells within the pancreatic islet cores. With age, the SMN mice gained more weight, had hyperinsulinemia, and increased hepatic glucagon sensitivity compared to the WT mice. The SMN mice, despite not having a SMA phenotype, still had profound changes in glucose metabolism with pancreatic abnormalities. In SMA human patients, disease severity is largely based on the SMN2 copy number; the more SMN2 copies present, the less severe form of disease.

The most recent Bowerman et al. study, although a mouse model, bridges the gap in understanding of the milder forms of SMA (types II, III, IV) and associated metabolic disease risk (13). A connection between reduced SMN protein, regardless of SMA phenotype expression, and pancreatic/metabolic dysfunction exists. Our study data highlight this unique metabolic dysfunction as well. The reduction in SMN protein results in a slow progression of glucose abnormalities, as seen in our 6 study subjects. Developing a screening protocol for clinical application, such as timed glucose and insulin testing for glucose tolerance, is the first step needed to better understand glucose metabolism and suspected metabolism abnormalities in a larger SMA population. An OGTT can provide these data and can be used as a way to screen patients and track metabolic changes.

All 6 of the study subjects were considered obese by National Health and Nutrition Examination Survey (NHANES) reference standards collected from 1999–2004 (24). Using DEXA scans, the NHANES data computed smoothed body fat percentages with corresponding population percentiles for children ≥8 years and adolescents (24).

The 95th percentile for boys (8 to 12.99 years) is 41.1 to 43.3%, while for girls it is 43.3 to 44.4%. Our subject DEXA scans reveal an average body fat percentage of 71.6% ± 13.1 indicating that the SMA subjects far exceed the 95th percentile for body fat. BMI calculations also indicate that 5 of the 6 subjects were overweight or obese based on BMI percentiles. Eighty three percent of the subjects were categorized as being overweight or obese (BMI ≥85th percentile) and 50% were considered obese (BMI ≥95th percentile).

Several studies have examined body composition for SMA patients and have found that body composition is not accurately depicted on a BMI growth chart (6, 9). Poruk et al. found that out of 47 type I SMA children, 68% of subjects plotted within the normal ranges, between the 3rd to 85th percentiles on the BMI chart. However, DEXA analysis revealed that the SMA children had substantially increased fat mass and lower fat free mass when compared to normal controls that plotted in the same BMI range. Sproule et al. reached a similar conclusion about body composition in SMA patients (9). Twenty-five children ages 5 to 18 years with SMA (2 type I, 13 type II, 10 type III) were assessed and compared to age-matched healthy children from NHANES data. Despite the SMA patients falling within the normal ranges on the BMI curve, DEXA analysis indicated a higher percentage of fat mass with diminished lean body mass. These higher percentages of fat mass placed them either in the "overweight" or "obese" categories. When using a receiver operator characteristic curve, BMI above the 75th, 50th, and 3rd percentiles corresponded to a FMI (fat mass index, fat mass/height in m^2) of >95th, >85th, and >50th percentiles, for the SMA patients. A high proportion of them also had a FMI above the 85th and 95th percentiles for age (9).

All SMA subjects had reduced insulin sensitivity as indicated by the large total sum of insulin values and calculated HOMA-IR values. A study by Kurtoğlu and colleagues studied insulin and glucose metabolism in 200 obese children and adolescents between the ages of 5 to 18 years (23). Based on their OGTT, a calculation of fasting glucose and insulin values (HOMA-IR) was applied and cut off criteria for obese children/adolescents was generated. The study results indicated that differences in the rates of insulin resistance were based on pubertal status. Those children in the prepubertal period, ≤8-9 years, had a rate of insulin resistance of 37% and 27.8% for boys and girls, respectively. The pubertal period, in contrast, was marked by a higher percentage of insulin resistance; over 60% for both boys and girls. They attributed this difference as a need for increased insulin resistance to aid in growth and development during puberty (23). HOMA-IR values for insulin resistance were calculated to be ≥2.67 and 2.22 for prepubertal boys and girls, while ≥5.22 and 3.82 was determined for pubertal boys and girls. Five of the 6 SMA subjects in our study met the criterion for insulin resistance based on their HOMA-IR values corresponding to their age. Interestingly, subject A, who had a glucose value of 169 mg/dL at the 120 minute interval during the OGTT, was not considered to have insulin resistance based on the HOMA-IR value. Our study results are in contrast with the Korağlu et al. study as the SMA subjects had a much higher rate of insulin resistance.

Body composition, obesity, and dietary intake have significant impact on the development of glucose intolerance for all populations (20). SMA subjects are at increased risk for developing obesity. Only one published case of type II diabetes in a SMA type II patient exists to date (25). However, our study results provide evidence that

as disease progresses, glucose intolerance and insulin resistance develops. Thus, it is imperative that dietary treatment be implemented to help control glucose and insulin values that were seen in our study. Research indicates that based on OGTT results, fasting glucose, 120 minute glucose, fasting insulin, and 100 minute insulin are associated with the highest rates of mortality later in life (26). In addition, a longitudinal study found a relationship between hyperinsulinemia preceding the development of type II diabetes 10 years earlier (27). Three stages for the development of diabetes have been proposed. The first stage includes hyperinsulinemia and normal to slightly elevated blood glucose. The second stage includes insulin resistance and glucose intolerance, while stage three is the development of diabetes. Many of the problems associated with the risk for cardiovascular disease (CVD) and metabolic syndrome occur during stages one and two, which occur 10 years prior to the development of diabetes (28). Consuming a diet that is consistent in complex carbohydrate, balanced with adequate protein, fiber, and reduced fat, is recommended for managing the progression of metabolic disease. In addition, awareness during times of illness and surgery, where dextrose may be administered for nutritional purposes should be considered. Increased amounts of dextrose could spike glucose and insulin levels, leading to episodes of hyperglycemia and the potential for rebound hypoglycemia as seen at the conclusion of the OGTT with subject E.

To address changes in body composition, strategies to preserve muscle mass and mobility should be considered. It is important that treatment for SMA patients involve working with an interdisciplinary team, which includes a physical therapist and dietitian. Many of our study patients did various types of physical activity, including, range of

motion exercises, pool therapy, dance, and standing in a support stander for extended periods of time. One subject (F) did conditioning and weight lifting exercise for upper body strength and to maintain lean muscle mass. This subject had the lowest percentage of body fat out of the group at 53.5%.

Strengths and Limitations

The strengths of this study include the diagnostic inclusion criteria and assessment methods. The primary goal was to understand whether glucose abnormalities exist in children and adolescents with SMA type II who are otherwise healthy without any prior diagnosis of glucose intolerance or diabetes. Furthermore, we took specific anthropometric measurements such as DEXA to further recognize the relationship between body composition and abnormalities with glucose regulation. In addition, all of the specific plasma measures taken throughout the test were necessary in the diagnosis of metabolic abnormalities and glucose intolerance by providing first time laboratory data in a controlled hospital environment. Overall, this study provides unique insight in a challenging population to recruit and study.

Limitations include the minimal number of participants, as sample size would not permit inferential statistical analysis. However, findings have provided benchmarks for establishing dietary management of glucose metabolism abnormalities. Although the 6 SMA subjects did not have age-matched healthy peers for study comparison, inclusion was not essential as normal ranges for the general population are well documented.

Conclusion

Our research provides novel evidence that glucose metabolism abnormalities exist for overweight/obese SMA patients. More research is needed on a larger scale to better understand the extent and potential treatments of such abnormalities. Further understanding will influence the nutrition therapy provided during the course of medical care, which may help in the prevention of metabolic disease.

APPENDIX: BASELINE AND OGTT CHECKLIST

Time Start	Task	Completed Y/N or record time
2 pm	Check into CCTS	
2:15-3:15 pm	Consent obtained	
3:15 pm	Vitals collected	
3:30 pm	Anthropometrics collected	
3:30-4:00pm	Clinical assessment	
4:00-5:00pm	DEXA scan @ School of Medicine	
5 pm	Standardized evening meal given	
8 pm	Standardized evening snack given	
10 pm	Overnight stay (8 hrs of fasting)	
6 am	Collect baseline urinary ketone for first void	
6:15 am	IV catheter placement, 20 or 22 gauge, with NS drip	
6:30 am	Baseline blood lab collection: -(hemoglobin A1c, IGF-1, blood glucose, insulin, glucagon, alanine, cortisol). Collect 0.5 ml for glucose analysis by YSI. Record glucose level in "Lab Collection Checklist".	
6:45 am	Preparation of glucose drink (1.75g glucose/kg body weight not to exceed 75 g)	
7 am	Glucose drink administered orally. Record time drink finished.	
---	Lab collection for: -(blood glucose, insulin, glucagon, alanine, cortisol)	
7:30 am	30 minutes	
8 am	60 minutes	
8:30 am	90 minutes	
9 am	120 minutes	
9:30 am	180 minutes	
	Collect 0.5 ml for glucose analysis by YSI. Record glucose level in "Lab Collection Checklist".	
9:45 am	Final urinary ketone collection	
10 am	Conclusion of data collection	
10:15 am	Meal of choice	

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